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# UNITED STATES ENVIRONMENTAL PROTECTION AGENCY WASHINGTON, D.C. 20460

JAN 4 1989

OFFICE OF PESTICIDES AND TOXIC SUBSTANCES

## **MEMORANDUM**

SUBJECT: 241-EUP-REN. Application for an Experimental Use Permit

to use AC 303,358 Chemical Hybridizing Agent on Cotton.

MRID Nos. 408064-01 and 408064-12. DEB No. 4485.

FROM: Linda S. Propst, Chemist

Dietary Exposure Branch

Health Effects Division (TS-769C)

THRU: Andrew R. Rathman, Section Head

Special Registration Section 1

Dietary Exposure Branch

Health Effects Division (TS-769C)

TO: Robert J. Taylor/James R. Yowell, PM Team 25

Fungicide-Herbicide Branch

Registration Division (TS-767C)

and

Toxicology Branch

Health Effects Division (TS-769C)

American Cyanamid Company is requesting an experimental use permit for AC 303,358, the potassium salt of 3,4-dichloro-5-isothiazolecarboxylate, a chemical hybridizing agent to use on cotton grown for breeding programs. This EUP request is for 2 years and all field trials are to be conducted in Maricopa County, Arizona. In 1989, 100 acres will be treated with a maximum of 180 lbs. acid equivalent (a.e.) in 1990, 250 acres will be treated with a maximum of 450 lbs. a.e.

Pennwalt Corporation submitted PP#6F3379 requesting that AC 303,358, also known as CHEMBRED, be exempted from the requirements of a tolerance. This exemption request was considered a food use has been transferred to American Cyanamid Corporation. Amercian Cyanamid Corporation requests that this EUP application be considered a non-food use.

## Conclusions and Recommendations

From the data submitted with this request, Dietary Exposure Branch concludes that residues of AC 303,358 will result in F1 cottonseed and in the F2 cotton plants.

Considering the limited size of this request, Dietary Exposure Branch recommends for this Experimental Use Permit. However, for full registration this use will be considered a food use and a tolerance will be needed on Fl cottonseed.

The registrant should be further advised that any deficiencies cited in connection with PP#6F3379 will need to be resolved before a tolerance can be granted.

## Proposed Use

AC 303,358 is applied foliarly at a rate of 0.1 to 0.125 lb ae/A early in the season when the first squares are observed. Repeat applications at 10-12 day intervals. At the sign of first bloom, observe flowers for male sterile, fertile, and female sterile flower parts. Increase the rate to 0.19 to 0.25 lb ae/A at the third application. Continue to observe new blooms for male sterile, fertile, and female sterile flower parts. The rate should be adjusted according to observations of the last bloom just before the next application. If significant male fertile flower parts are present, increase the rate by 0.1 lb. a.e./A. If excessive sterility is observed, reduce the rate by 0.1 lb Increase rate to 0.25 - 3.0 lb a.e./A at the fourth application. Continue applications about every 10-12 days adjusting the rate or timing as needed until two weeks prior to the bloom which is expected to produce open bells. The maximum total application per season should not exceed 1.8 lb. a.e./A.

Seed from treated plants must be kept separate and may be used for planting purposes only. Treated fields are to be planted only to cotton grown for seed the following year.

#### Plant Metabolism

## Phase 1

Four cotton plants, 18 to 24 inches high with pinhead squares showing, grown in a greenhouse in individual six inch diameter pots were used in Phase I of this study. Two plants were treated by side dressing (placing the pesticide in the root zone about two to three inches from the stalk at a depth of two to three inches) with the equivalent of 1 lb.a.i./acre of <sup>14</sup>C 3,4-dichloro-5-isothiazole carboxylic acid (DICA). In addition, the equivalent of 50 lbs. per acre of prilled urea was incorporated into the soil at time of treatment.

The remaining two cotton plants received three foliar sprayings with the total applications equivalent to 1 lb. a.i./acre of  $^{14}\text{C}$  DICA at 7 - 8 day intervals with the last treatment applied approximately 8 weeks prior to harvest. The

prilled urea was also incorporated into the soil of these plants at the rate of 50 lbs. per acre.

All plants were individually isolated to contain the radioactive materials and watered as necessary to maintain growth. Bolls were harvested approximately 4 months after planting.

## Phase II

For this phase of the study, six 18 - 24" plants were used. The plants were placed in individual square (1.3 foot) trays. The soil in each tray was mounded to simulate a row/furrow. The plants were divided into three groups of two for treatment.

Two plants received three foliar applications of 1.0 lb. ai/acre <sup>14</sup>C DICA at 7 day intervals. Urea prills were incorporated into the soll at the rate of 50 lbs./acre.

Two plants received a side dressing of urea (50 lbs./acre) impregnated with  $^{14}\text{C}$  DICA. The urea prills were mixed in a vial with 1 lb. a.i./acre of  $^{14}\text{C}$  DICA and allowed to dry overnight. The dried urea prills were incorporated into the soil at a depth of 3-4 inches approximately 3-4 inches away from the stalk.

Attapulgite granules were impregnated with <sup>14</sup>C DICA at the same rate and fashion as the urea prills above. The plants were side dressed with the Attapulgite granules. The 50 lbs./acre of urea was incorporated into the soil.

Ten days following the third foliar application made to the first two plants, all plants were flooded by filling the container with sufficient water to nearly cover the "hill" as would be the case in a cotton field. The water was siphoned off about 16 hours later. The second flood watering took place 9 days later. Prior to the "floodings", the plants were watered only to maintain vigorous growth rather than attempting to simulate in-furrow irrigation in the field.

#### Phase III

Six cotton plants ranging from 18 to 24 inches were planted and maintained with the addition of urea the same as the six plants received in Phase II. However, the treatment rates of  $^{14}\text{C}$  DICA were altered to 3/4 lbs. a.i./acre total. The plants receiving the foliar applications received 5 applications at 0.15 lb. a.i/acre of  $^{14}\text{C}$  DICA. The plants were initially flooded five days after the first foliar application or side dress applications. After three days the excess water was siphoned off since the level was remaining essentially the same except for a minor amount of evaporation. The plants were flooded six more times by filling the trays to within two to three inches of the

top of the hills, allowing the water to stand about six hours and then siphoning away the free water. Bolls designated as P3S1, P3S2, P3F1, P3F2 (one boll each) were harvested 3 weeks after the last flooding. Three weeks later bolls were harvested from plants P3S1 (3), P3S2, P3F1 (2), P3F2.

Six weeks after the last flooding a boll was harvested from plant P3F1. Nine week after the last flooding a boll was harvested from plant P3S2.

## Phase IV

Six cotton plants identical to the ones used in Phase III were treated at the same rates as the plants in Phase III. The plants were watered (6X) by flooding for six hours and then siphoning off the excess water. Bolls were harvested from plants designated P4F1, P4F2, and P4S2.

# Growing F2 Plants for Residue Determination

Seeds (F1) from treated plants described in Phase I - IV of known DICA content were planted in individual pots in the greenhouse. In addition, control seeds from a commercial source were planted. All seeds were allowed to germinate and grow. The plants were kept watered and under light conditions conducive to optimal growth. The temperature was kept elevated as is common in the cotton growing regions. Entire plants were taken to determine the DICA levels at 1 true leaf stage, 4 true leaf stage, 6 true leaf stage, pinhead square stage, and last sample 4 to 7 days pre-bloom.

At each sampling, one check and two or more treated plants were taken. The plant stem was cut off at soil level so that no part of the root system was included in the sample.

## Production of F2 Seed

Seeds obtained from the treated plants in Phase I - IV were planted in individual pots and grown in the greenhouse. The plants were maintained throughout by watering and fertilizing with general purpose fertilizer to promote normal growth. The ripe bolls from these self-pollinated plants were harvested after they spilled their lint. Each boll was hand ginned and the resulting F2 seed set aside for future analysis.

#### Analyses of DICA in Fl and F2 Cottonseed and in F2 Cotton Plants

Whole individual F1 and F2 cottonseeds were weighed and wrapped securely in Whatman No. 29 ashless paper. The samples were sealed in a Thomas-Ogg flask coated with hyamine hydroxide and flooded with oxygen. The samples were combusted in a

Thomas-Ogg apparatus and the  $^{14}\mathrm{CO}_2$  allowed to be absorbed in the hyamine.

After a suitable absorption period, scintillation cocktail was added to the flask and the contents mixed. An aliquot of the solution was pipetted into a counting vial and the radioactivity determined by liquid scintillation counting.

Whole plants were weighed and homogenized with methanol. The homogenate was transferred to bottles and centrifuged. The clear supernatant was decanted and collected for liquid scintillation counting. The pellet was re-extracted with methanol. The homogenate was filtered. The filter cake was washed with additional methanol.

The filtrate was combined with the supernatant and reduced in volume to 25 ml. Aliquots were mixed with Aquasol and the radioactivity determined by liquid scintillation counting.

The filter cake was allowed to air dry thoroughly for subsequent combustion. The dried extracted plant material was weighed into Whatman No. 29 ashless paper, combusted, and counted in the same manner as the cottonseed.

## Results

Residues of DICA in F1 cottonseed ranged from 6.5 ppm to 93.9 ppm. These results are the average of duplicate analyses of seed from a single boll. The lowest residue was found in the P4S2 seed and the highest residue in the P2SU1 seed.

No residues were detected in F2 cottonseed.

Total residues of DICA in F2 cotton plants are as follows:

Plant "	lst TL 16 Days	4th TL 22 Days	6th TL 31 Days	Pinhead Squares 45 Days	Final 62 Days
	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)
Check	<0.01	<0.01	<0.01	<0.01	<0.01
P4F2	1.20	.56	.13	.01	.03
P3F1	2.31	. 76	.09	<0.01	-
P3S1	-	.69	-	-	.02
P3S2	_	-	-	-	<0.01

TL=True Leaf

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From these studies we can conclude that residues of AC 303,358 will result in the F1 cottonseed and in the F2 plants. No attempts were made to characterize or identify the residue found in the F1 cottonseed or the F2 cotton plants.

In light of the limited size of this Experimental Use Permit, Dietary Exposure Branch will consider this a non-food use. However, for a full registration in the future this use will be considered a food use and a tolerance will be needed on F1 cottonseed.

cc: Reading File, Circulation, Subject File, Reviewer, PP#6F3379, PMSD/ISB RDI: A. R. Rathman, 1/3/89; R. D. Schmitt, 1/3/89 TS-769C:DEB:LSP:lsp:CM-2:Rm803-C:557-7324:1/4/89

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